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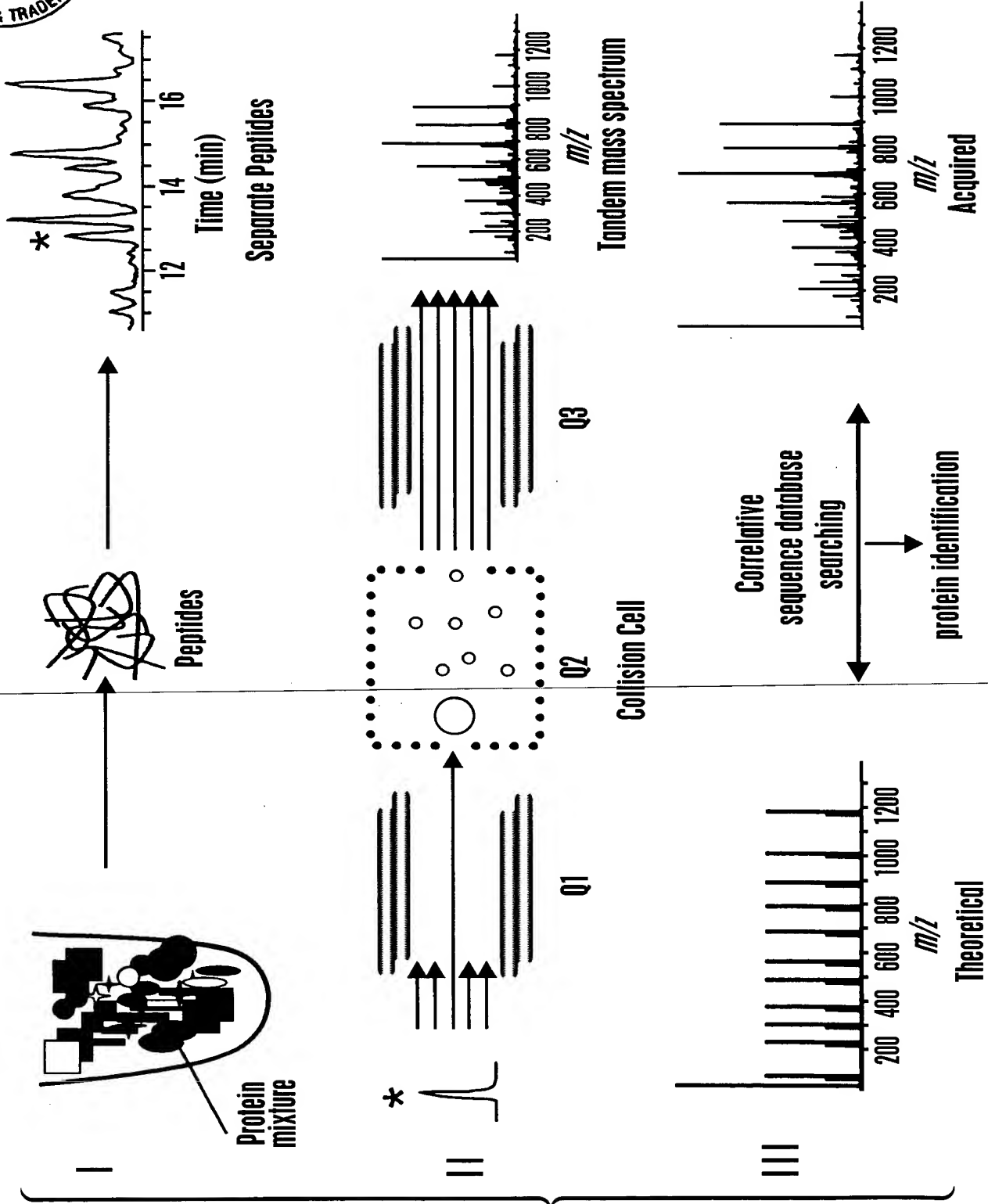
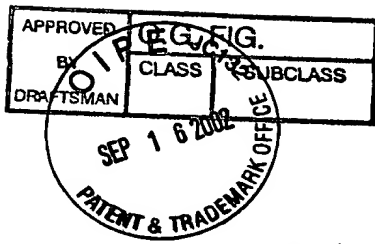


Figure 1

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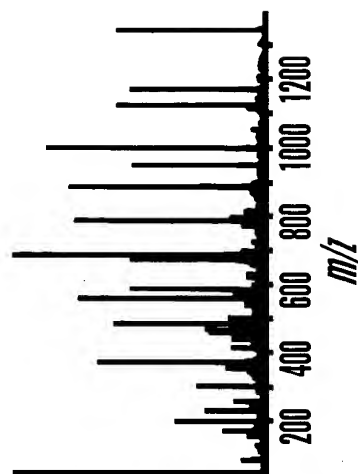
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APPROVED	O.G. FIG.	
BY	CLASS	SUBCLASS
DP/AF/TS/MA		



Mass spectrum consists of fragment ions specific to selected parent ion.



Mass spectrum consists of fragment ions from multiple parent ions.

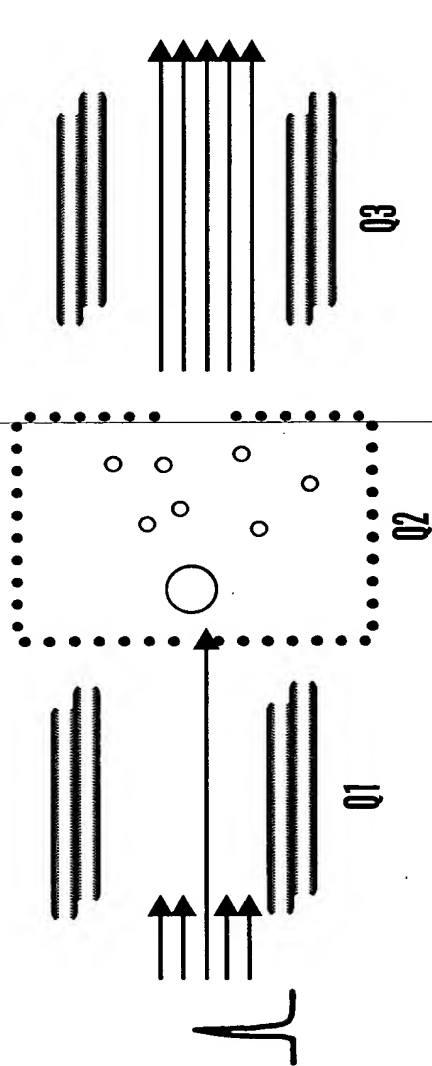


Fig. 2A

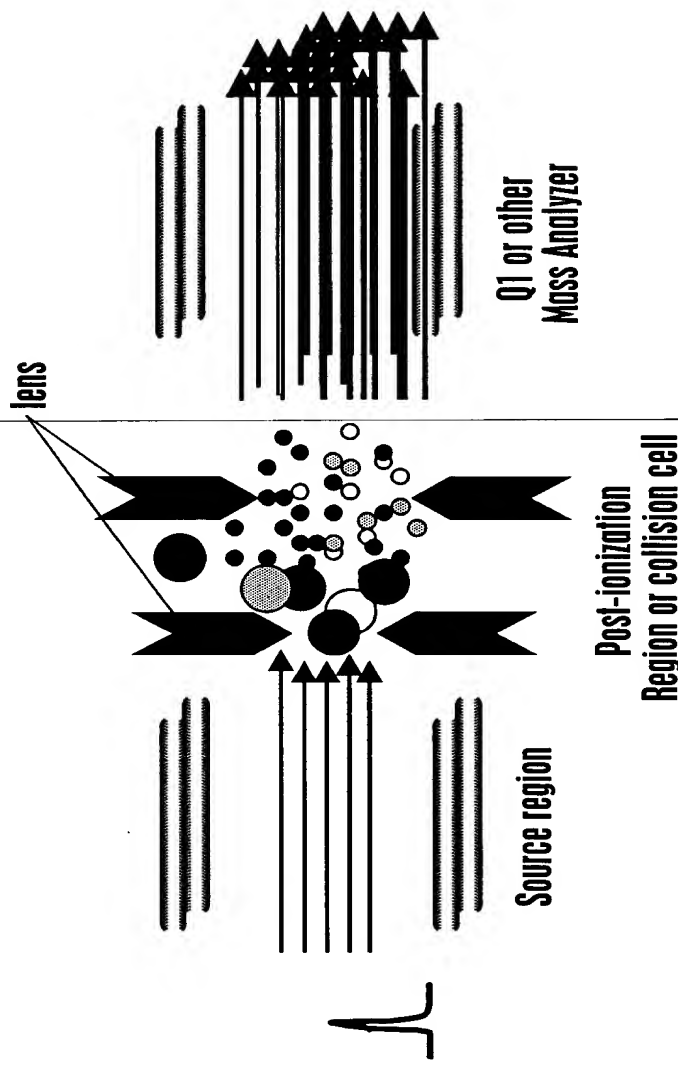


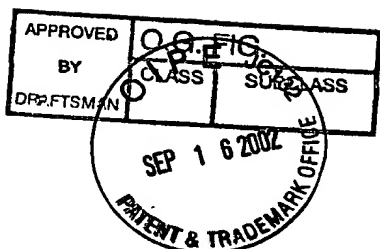
Fig. 2B

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**Growth Condition 1**

1a) Grow Cells or Harvest Tissue from Biopsy or Collect serum or spinal fluid, etc.

Growth Condition 2

1b) Grow Cells or Harvest Tissue from Biopsy or Collect serum or spinal fluid, etc.

State 1

2) Harvest Proteins & Label separately with Isotopically distinct ICAT reagents such that State 1 & 2 can be distinguished after mixing.

State 2**State 1****State 2**

3) Combine labeled Proteins from States 1 & 2.

1 + 2

- 4) Fractionate proteins by molecular weight.
- 5) Digest proteins to peptides (e.g. using Trypsin).
- 6) Separate peptides by ion exchange and collect fractions.
- 7) Purify each ion fraction by affinity chromatography.
- 8) Analyze each affinity chromatography fraction by LC/MS or MALDI-MS after LC fractionation.

9) Use annotated peptide tag database to identify proteins and quantitate relative abundance between states.

Figure 3

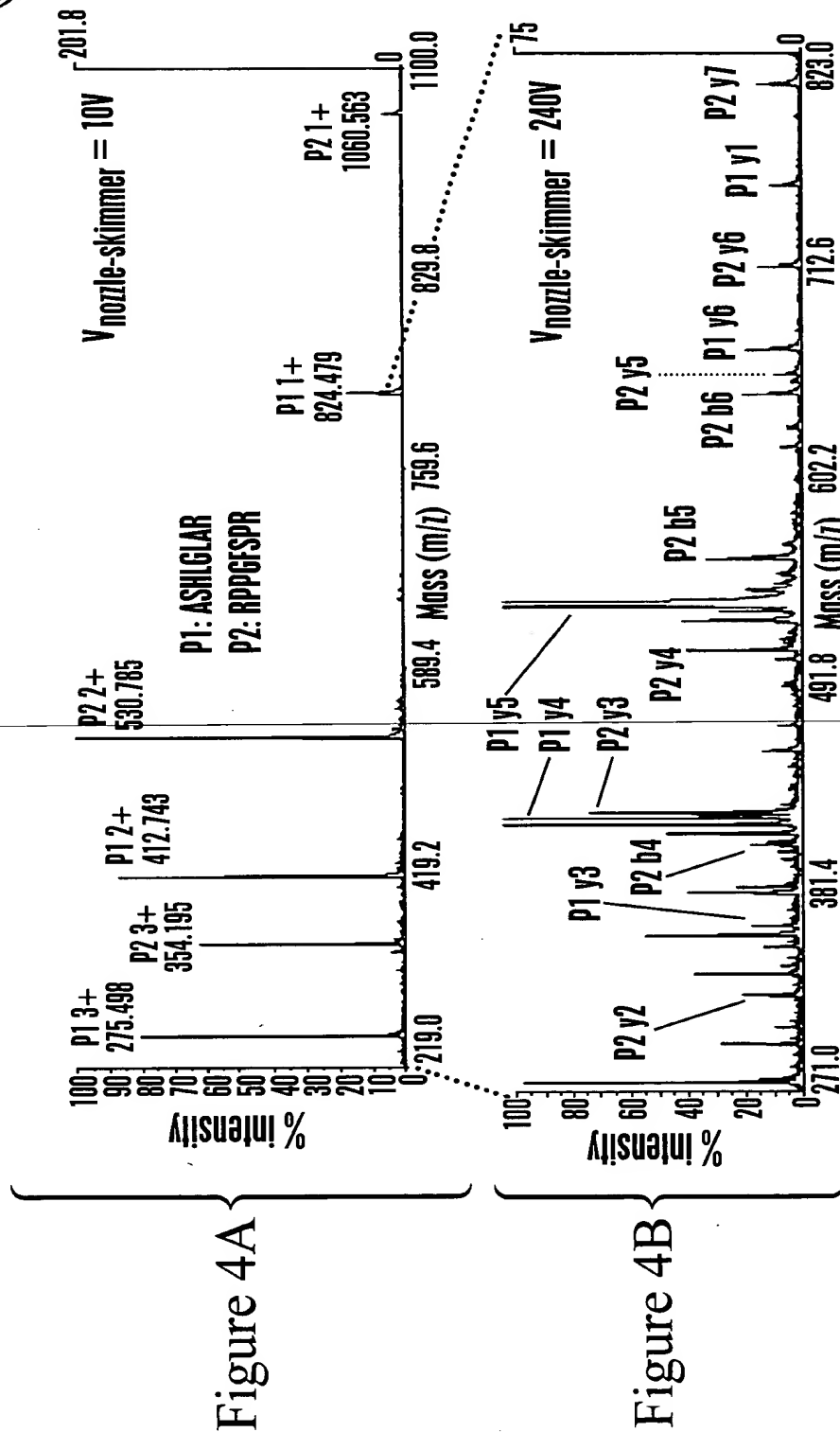
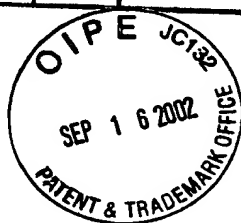
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APPROVED	O.G. FIG.	
BY	CLASS	SUBCLASS
DP,FTSMAN		



APPROVED	O.G. FIG.	
BY	CLASS	SUBCLASS
DRAFTSMAN		

Title: Rapid and Quantitative Proteome Analysis and Related Methods
 Inventor: David R. Goodlett
 Serial No.: 09/748,783 (Confirmation No. 3333)
 Filing Date: December 26, 2000
 Attorney Docket: P-IS 4369
 USPTO Customer No. 23601 (858-535-9001)

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Human protein database:

60,884 sequence entries

Fragment ion masses used for analysis:

710.385
 753.435
 807.374
 886.402
 904.451
 1001.564

 Bradykinin RPPGFSPFR MH+ 1060.5676

at 10 PPM, 144 tryptic peptides match

13 matches using fragment ion constraint:

ACISEILPSK
 GVRYSFGFK
 RANLISQCR
 RCGLPSSGKR
 RDITLEASR
 RERETLEK
 RLTEEERK
 RLVEVDSSR
 RNLLDHR
 RPHAAQPGAR
 RPPGFSPFR **
 RPQTATASTK
 RRPSAYQAL

 ASHLGLAR MH+ 824.4739

at 10 PPM, 57 tryptic peptides match

12 matches using fragment ion constraint:

ACYIKVK
 ADPLPRR
 AFVAFAAK
 AFVFGRK
 AHAEIRK
 AHEAKIR
 ALEAHKR
 ALQFFAK
 AMAIYKK
 APDPRLR
 ASHLGLAR **
 AVAGHLTR

Figure 4C